MARINE BIOTECHNOLOGY IN THE NORTHEAST REGION:

Current and Future Research Initiatives

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Report on the Northeast Regional
Sea Grant Collaborative Workshop
Held on December 1, 1993
Massachusets Institute of Technology

Sea Grant College Program
Massachusetts Institute of Technology
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INTRODUCTION

On December 1, 1994, leading researchers from throughout the northeast region gathered at to MIT attend the Northeast Regional Sea Grant Collaborative Workshop, "Marine Biotechnology in the Northeast Region: Current and Future Research Initiatives." This publication reports on the discussions and conclusions reached at this workshop.

The primary impetus for this workshop was the recent passage by Congress of the "Marine Biotechnology Investment Act." This act appropriated $3.2 million in fiscal year 1994 for enhancing the National Sea Grant College Program's efforts in marine biotechnology, by supporting Sea Grant efforts in research, education and outreach.

The workshop brought together over 60 researchers from throughout the northeast region to discuss current issues in marine biotechnology research and identify key areas of interest for future research efforts. By dividing the workshop into 5 break-out groups, the attendees were able to focus on specific theme areas for discussions. These theme areas were: biotechnology research for the aquaculture industry and fisheries management, bioactive materials for industrial and food processes, marine biomaterials for medical applications, biomaterials for sensor and probe development, and advisory and educational program initiatives. The findings of these groups are presented within this report.

Another objective of this workshop was to provide an environment where would have the opportunity to establish important links among themselves, which may lead to collaborative research proposals for funding under the marine biotechnology strategic research initiative. This indeed did occur with the MIT Sea Grant Program receiving 9 marine biotechnology proposals, several of which were collaborations among researchers that attended the workshop. For example a major research proposal has been submitted which, if funded will be a collaboration between MIT, the University of Maryland, Brown University, the City University of New York, and Harvard. Another collaboration, for an advisory/education proposal brought together MIT, the University of Connecticut, Harvard, and the University of Massachusetts.

It is our hope that as future strategic initiatives are identified within the Sea Grant Program, workshops such as the one MIT recently organized will take place. The workshop format provides a unique opportunity to bring researchers together to identify the important questions that require solutions and ultimately establish the connections that will lead to future collaborative research efforts.

John Moore, Jr.
Workshop Coordinator
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WORKSHOP AGENDA

Northeast Regional Sea Grant Collaborative Workshop

Marine Biotechnology in the Northeast Region:
Current and Future Research Initiatives

Massachusetts Institute of Technology
December 1, 1993

9:00	**Welcome & Introduction** *Chrys Chryssostomidis, Workshop Co-Chairman,* *MIT Sea Grant College Program*
9:15	**Discussion of Workshop Format, Goals & Objectives** *Discussion Leader: Robert Langer, Workshop Co-Chairman, MIT*
10:30	**Break-Out Discussions** Group 1: Biotechnology Research for the Aquaculture Industry and Fisheries Management *Group Leader: Yonathan Zohar, University of Maryland* Group 2: Marine Biomaterials for Industrial & Food Processes *Group Leader: Herbert Hultin, Univ. of Massachusetts* Group 3: Marine Biomaterials for Medical Applications *Group Leader: Robert Langer, MIT* Group 4: Biomaterials for Sensor and Probe Development *Group Leader: David Walt, Tufts University*
12:30	**LUNCH** *Keynote Speaker: John Burris, Director,* *Marine Biological Laboratory*
1:45	**Continue Break-Out Discussions of Groups 1-4** **Begin Group 5:** Advisory and Educational Program Initiatives, *Group Leader: John Moore, MIT*
3:30	**Presentations by Break-Out Session Leaders of Group Findings**
4:15	**Wrap-up Discussions**
5:00	**ADJOURN**

BREAK-OUT SESSION REPORTS

Biotechnology Research for the Aquaculture Industry and Fisheries Management

The work group discussed research directions related to the aquaculture industry and to the understanding and management of wild populations of marine organisms. Basic research on the molecular, cellular and physiological aspects of the following areas were recommended. Due to the large size of this group a broad range of research needs were identified and are listed below. Of these many research needs, the group focused on four specific areas that the core competencies of the northeast region could address.

Reproduction, Development and Growth

To improve both the production of aquaculture species and maintain existing wild stocks of fisheries, much more needs to be understood of the basic reproduction, development and growth functions of marine organisms. Presently, little is known about the optimal nutritional requirements at the different life stages (larvae, grow-out, fish, broodstock). An understanding of these requirements will lead to an increase in the efficiency for feeding aquaculture stocks and will assist in the development of more effective fisheries management plans. In addition to nutritional requirements, the marine biota's means of adaptation is also an area of interest. The methods of adaptation, to the environment (photoperiod, temperature, salinity, pollutants, etc.) and its potential effect on an organisms reproduction and growth may assist in tailoring and optimizing farmed organisms to different environmental conditions.

Future research required on the environmental, hormonal and molecular control of reproduction, development and growth. With reproduction particular areas of interest include: study of gametogenesis, egg quality; maternal effects; stress-reproduction interactions, technologies for the control of sex differentiation, the onset of sexual maturity and the induction of spawning year-round. In development and growth of marine organisms, areas of interest are: ontogeny of the digestive and immune systems, osmoregulation, technologies to increase survival rates and success of metamorphosis, gene transfer for growth hormones and growth factors, growth-reproduction interactions, effects of stress, and techniques to enhance growth rates and optimize flesh quality.

Disease

There is a need for further study of fish immune systems. These studies should include: stress-immune response interactions, host-pathogen interactions, and genetic selection; transgenesis for disease resistance. Future research efforts are also needed to develop a number of techniques that include: in-vitro cultures for disease studies, sensitive probes for efficient diagnostics, efficient vaccines and vaccine delivery methods, and develop efficient therapies for treatment of diseases.

Drug Delivery

New drug delivery systems have broad applications in the aquaculture industry, both as an efficient and potentially more effective way to administer a wide variety of drugs that include among others: hormones, vaccines, therapeutics, nutrients, and vitamins. Further study is needed to understand the various uptake mechanisms and their potential impact, to reproduction, larval development, growth, nutrition and disease.

Genetic Selection and Diversification

Genetic selection techniques could be developed to identify different traits that will improved reproduction, survival, growth, disease resistance, etc. The use of cryopreservation of gametes and embryos and the development of gene banks should also be explored. Diversification studies include the introduction of new species that may be of interest for the seafood market and organisms that may be sources of innovative biomaterials such as polymers, enzymes and pharmaceuticals.

Stocks and Species Identification

Using molecular tools for *in situ* identification of species with conservative morphologies, for sorting novel unknown species and for understanding population kinetics of larvae of species which are important for fisheries.

It was decided that in light of the needs of the aquaculture and fisheries industries and of the scientific expertise in the Northeast region, the following four areas should be prioritized within the first round of the Marine Biotechology initiative:

Novel Technologies for Delivery of Regulatory Biomolecules

In order to make aquaculture in the Northeast a more intensive, efficient and profitable industry, there is a need to control reproduction of the farmed organisms (year-round spawning, advance or delay puberty, manipulate sex differentiation), to increase the success of their early development and metamorphosis, to accelerate their growth rate and to enhance their resistance to disease. Achieving these goals relies on understanding these processes and their regulation and on the capability to efficiently administer into the organisms biomolecules such as hormones (reproductive hormones, growth hormones, growth factors) and therapeutics (vaccines, antibiotics). The basic interactions between these molecules and the processes that they modulate (reproduction, development, growth and immune protection) should be explored, as well as the most efficient patterns of administration (pulsatile, sustained, etc.) leading to enhancement of aquaculture organisms. On the basis of this information, the molecules are incorporated into the appropriate polymer-based controlled-release delivery systems that will release them in the optimal pattern. The delivery systems will use either oral or parenteral (injection or via the water) routes. The kinetics of uptake of the biomolecules by the studied organisms will be monitored. The effects of the biomolecules on the functions of interest (reproduction, growth, development and immune protection) will be explored. Optimal delivery systems for increasing the performances of farmed organisms will be identified and recommended for commercial uses.

Problems in the Area of Disease of Aquatic Organisms

With the intensification of aquaculture and the increase of pollution in major coastal areas, disease emerges as a major problem to both aquaculture and fisheries industries. In recent years a virtual explosion of disease-related technology based on molecular biological techniques in mammalian systems has occurred. Transfer of this technology to marine systems is very slow. Part of the problem is determining how to make the transfer to lower vertebrates and invertebrates. It is proposed that this could best be done through a comparative approach of defining basic conserved molecular mechanisms. In order to do so, an invertebrate (shellfish) and a lower vertebrate (finfish) model are developed contributing to our basic understanding of immune and disease mechanisms and the development of diagnostics and vaccination methods. The studied models chosen are ones that are of importance to the aquaculture or the fisheries industry. It is proposed that diagnostic and vaccination tools be developed using recombinant DNA techniques to detect the presence of disease and overcome it. Efficient diagnostics are proposed to screen both natural populations and cultured stocks before the populations are severely infected.

Environmental Concerns
Intensive aquaculture should be harmless to the environment and to the consumers. Research in this area will focus on three major objectives:

Development of identification and detection techniques for marine species of aquacultural importance:

- Marker ID techniques for hybrid species to be used in the animal husbandry of aquaculture species (tilapia, striped bass).
- Track introduced species of fish and aquatic plants.
- Detect human pathogens in seafood.
- Develop database of existing detection/ID methods and provide access through national computer networks.

Address concerns about effluent from intensive aquaculture methods:

- Develop control methods.
- Establish scientific basis for regulations.
- Develop analytical methods to monitor chemical, microbial and biochemical parameters.
- Understand biological filtration and develop closed "environmental friendly" aquaculture systems.

Develop strategies for removal of microbial pathogens and other environmental contaminants from seafood species.

Stock and Species Identification
Research in this area will focus on two major objectives:

Understanding larval dynamics; identification of individuals in plankton samples; evaluation of biodiversity.

- Develop techniques to sort and identify key plankton species in situ. For example, one promising technique is whole cell *in situ* PCR with fluorescent probes. This technique may provide for the precise and reliable enumeration of larval supply for recruitment of fisheries.
- Sorting of plankton samples for novel unknown organisms. Benthic samples may have short larval stages where they can be captured. The use of molecular techniques will enable researchers to identify organisms with very conservative morphologies (molluscs, worms, crustaceans) and detect new species. It will allow detecting true levels of diversity and could be exploited to compare disturbed/undisturbed areas or ecological systems.

Study historical processes in commercially and ecologically important taxa by examining samples of scales or tissues.

- Population/species-specific PCR markers will provide variability. This can be used to (a) understand the genetic reaction of populations to exploitation, (b) evaluate models of population dynamics and (c) study historical trends in contaminant levels causing mutations.

Marine Biomaterials for Industrial and Food Processing

The group charged with developing the potential of marine biomaterials for industrial and food processing suggests three broad areas of concern, namely: environmental protection, economic development, and consumer safety.

In the area of environmental protection, major concerns are the loss or impending loss of economically important technologies that are no longer environmentally acceptable. This includes the use of tributyltin as an antifoulant and the use of aluminum and ferric salts for waste water treatment. Their is also a need for more effective bioremediation technologies for managing marine pollution. Marine biotechnologies hold promise of environmentally acceptable and economically beneficial treatments for all of these current needs.

In the area of economic development, Northeastern fishing communities have been devastated by the declining stocks of cod, haddock and other traditional species. Jobs have been lost in fishing, fish processing, and transport while U.S. fishery imports (second only to petrochemicals in non-manufactured goods) soar. Marine biotechnology holds self-evident promise here, offering tools for the culture and capture of new and underutilized species, their processing into human food products, and the more complete utilization of all catches. The group was particularly concerned by the fact that more than half of the seafood harvested is disposed of as garbage or very low quality product; the up-grading of the rest of the catch and the development of high value products from it are of great importance.

Consumer safety is a major issue. Consumers are warned about seafood dangers almost as often as they are told that seafood is "heart-healthy." In the Northeast, the shellfish industry is the greatest casualty of this concern, with consumers learning of bed closures, outbreaks of illnesses, and illegal harvests. While we recognize the urgency of the need for consumer safety assurance for shellfish, we also believe that better quality assessment of finfish will aid the industry inproducing a higher value, more consistent product.

Environmental Protection

Many marine biotechnologies have potential applications in the field of environmental protection. The problem generally stated is that new technologies with more acceptable environmental impacts need to be developed to replace those technologies that are no longer acceptable. The justification for enhanced funding in this area is that previous control technologies or their absence has led to persistent, far-reaching (detrimental) effects on environmental quality. Novel marine biotechnological solutions should be exploited to develop new control technologies for coastal and marine facilities. For example, marine algae, bacteria, or by-products from any number of organisms could be used to clean up the waste streams of coastal industry and food processing plants. Bioreactors or holding ponds with selected or engineered marine organisms capable of degrading or concentrating contaminants offer many possibilities. Another example is the use of natural products, e.g., secondary metabolites, as antifouling agents on marine installations to replace toxic metal coatings. Toxic metals used in antifoulants currently accumulate in harbors and cause chronic habitat degradation. Bioremediation of existing contaminated marine environments also requires further research in the development of "engineered" organisms.

Economic Development
Great opportunities exist for economic enhancement and development via marine biotechnological research and development. The fishing and fish processing industries in the Northeast region of the U.S. have been directed towards a few species. Overfishing, poor management and environmental pollution have drastically reduced these traditional species to the point where the industry is on the verge of collapse. Several approaches are needed to reverse this trend. First, the suitability of a much wider variety of species for human food and other uses must be determined; this would include finfish, shellfish and algae. These organisms could also be sources of enzymes, drugs, antioxidants, pigments or other compounds with unique properties suitable for specific uses. The development of markets for these currently underutilized species would assist this troubled industry. In addition, improvement of valuable species is needed. This might include biotechnological approaches to reduce the period of "soft-shell" in lobster and improve roe production in sea urchins.

The flesh of traditional species is poorly used; up to 50-60% remains on the carcass after filleting. This should be recovered, and the processing of non-traditional species should include process development to prevent this waste.

Unlike the situation with land animals and birds, little genetic improvement in properties as food has occurred with seafood over the millennia. The tools are now at hand to correct this. Genetic manipulation and engineering of finfish, shellfish and seaweeds will be an important aspect of the future development of a healthy, thriving seafood industry that reestablishes economic vitality in coastal areas, provides essential nutrients to the U.S. consumer, and allows the U.S. to compete in an ever-increasingly competitive world market.

Consumer Safety
Issues of safety and quality of seafoods are becoming an ever greater concern to consumers and industry alike. Contamination problems have necessitated the closing of shellfish beds all along the East coast. In addition, our ever increasing knowledge of tissue component breakdown and their relation to quality and economic value of seafood products can be exploited by development of biotechnological assays to objectively determine seafood quality and economic value. The extraordinary possibilities and unique selectivity and sensitivity introduced by recombinant DNA techniques and the use of biological molecules such as monoclonal antibodies and enzymes for detection of potentially harmful microorganisms and chemical constituents, all important in seafood safety, and quality will produce results previously unattainable.

At a time when people are becoming more aware of not only the value but also the potential hazards of consuming seafoods in various forms. The advances possible in marine biotechnology presents a unique opportunity to both guarantee the quality and safety of these important resources. Hopefully, Sea Grant will rise to this challenge and become a leader in developing the remarkable opportunities available.

Marine Biomaterials for Medical Applications

The group focused on three specific areas where research and development efforts, supported through the National Sea Grant Marine Biotechnology Initiative, would have potentially broad and significant impacts in the medical field. The three areas identified are, bioadhesion and metal ion sequestration, drug delivery systems, and new drugs and antibiotics.

Bioadhesion and Metal Ion Sequestration

Marvelous feats of engineering are accomplished by the diverse organisms living underwater. Our technology cannot yet accomplish the mussel's feat of producing a strong and flexible tethering cord that can also serve as its own glue, and do so whether underwater or exposed to the atmosphere. Research into such spectacular feats has disclosed new examples of interesting compounds in this and other marine organisms. A specific example that could be investigated is the hydroxylated ring structures that appear to play important roles in underwater adhesion, healing, and metal ion sequestration.

Bioadhesion

It is widely recognized that marine organisms, such as the New England blue mussel (*Mytilus edulis*) and the barnacle, are now joined by the fresh-water immigrant *Driessena polymorpha* (the zebra mussel) in being Nature's most effective underwater adhesion specialists. Studies of their remarkable abilities to attach to already slimy surfaces of almost all types, and of the few materials that resist such attachments, can go a long way toward helping humans better engineer the molecules needed for promotion or prevention of bioadhesion in a similarly large range of circumstances. The availability of test organisms and bioadhesive substances spanning the entire range of salinities, from the fresh water Great Lakes to the saltiest oceans, is a unique benefit to this aspect of biotechnology research, since the most interesting range of salinities -- that of human physiology (saliva, synovial fluids, blood) -- is between that of fresh and ocean waters! Thus, one confident prediction of a beneficial outcome from this research will be the development of safe and effective ways to secure mucosal adhesion for drug delivery devices. A more speculative outcome will result from the fundamental understanding of the DOPA (*dihydroxyphenyalanine*) moieties of Nature's bioadhesive glycoproteins. DOPA is also involved in understanding various human conditions from Parkinson's disease to pain control. These special hydroxylated ring structures, already implicated in interfacial dehydration as the first step in bioadhesion and as crosslinkers for the adhesive itself, also seem to be involved in forming the underwater protective sheaths of various organisms' attachment structures, rendering them free from biodeterioration by infective microorganisms and other environmental challenges.

Metal Ion Sequestration

Involvement of related hydroxyl-ring structures in metal ion binding also promises to be important in preventing metal ions from migrating from implantable bioprothesis. For example, the metal vanadium has long been prized as an important additive to steel that improves its strength and durability. These attributes have led to its inclusion in biomedical devices such as dental implants and artificial joints. As these devices perform their functions in daily living, they wear down and the vanadium diffuses into the human body. How better to understand the physiological chemistry of such a potentially harmful intruder, than to study the role of this metal in a marine organism that naturally and specifically accumulates vanadium. The common sea squirt selectively sequesters vanadium from among the dozens of more highly concentrated metal ions in the marine environment and accumulates the metal ion in certain blood cells at concentrations more than 10 million times the concentration in sea water. In the course of investigating this phenomenon, a new compound -- dubbed tunichrome after the scientific name of the sea squirt, tunicate -- possessing three hydroxylated benzene ring systems was discovered. It would be useful to understand whether the compound is used in sequestering vanadium, whether it is there together with vanadium to provide resistance to bacterial infection, or whether it assists in the rapid suturing of wounds inflicted by predation.

Drug and Cell Delivery Systems

Drug delivery is one of the most important problems in medical science and in a short time, annual sales exceeds $4 billion. New types of delivery systems and the expansion of delivery systems to marine problems could greatly expand these opportunities.

Marine biomaterials such as alginates, carageenans, chitin and others may be very useful as components of drug delivery systems. These substances have been shown to be safe in the human body and because they can be used in conjunction with mild encapsulation methods, they are capable of encapsulating sensitive molecules such as proteins. By adjusting various material properties, they may be able to release molecules in a sustained manner for long periods of time.

One interesting type of marine biomaterial are alginates. Alginates possess the unique property that they can be placed in water or injected into an aqueous bath of divalent or trivalent cations and gelled instantaneously. This provides one of the mildest encapsulation procedures known is successfully used, for example, to encapsulate mammalian cells such as islet and many other mammalian cell types. This encapsulation allows mammalian cells to secrete important molecules such as insulin. Thus, the alginate provides a vehicle for protecting the cells from immune destruction by the body. Yet, it still enables the cells to deliver the desired components which may be useful in the treatment of diabetes and other diseases.

Although the above materials may be useful biomaterials, they have certain limitations. When developed as carriers for low molecular weight therapeutics, small molecules easily diffuse out of the delivery system. There is a need to develop novel bioerodable biomaterials in order to circumvent some of these problems. Polymers such as polylactide co-glycolide and polyanhydrides are needed since they can be tailored to degrade form within several days up to several years. In addition, there is a need to develop new technologies to make injectable or orally administered drug delivery systems based on microspheres. They should be designed with the notion that the drug should be protected from the environment and simultaneously allow controlled degradation and release of the drug. Some useful procedures for making microspheres are spray drying, solvent evaporation, solvent removal and so on. These types of systems should, with further study, be very useful as delivery systems for nutrients, vaccines, hormones and other entities to fish and aquaculture; as delivery systems for agents to protect sea life or to destroy unwanted entities, (e.g. agents to combat water borne parasitic diseases such as schistosomaisis).

New Drugs and Antibiotics

The search for new marine-organism-based pharmaceuticals, medical reagents and tools from marine sources for medical research or therapeutics has already yielded successful products and novel applications. These products have become important precisely because of the uniqueness of the marine environment and organisms. Examples of some products or applications include protein phosphatase inhibitors from blue-green algae and sponges, which have no counterparts in terrestrial organisms. Several macromolecules, such as lectins from marine macroalgae, are used in blood typing, cancer research and immunological research. These lectins are very different in terms of their carbohydrate binding specificities than lectins from higher plants in that they recognize complex carbohydrates. Endotoxin binding proteins for the horseshoe crab are widely used in the health care industry for diagnostic purposes. Several natural products such as bryostatin are now in clinical cancer trials and may become new drugs.

Much of the success in identifying new marine-derived pharmaceuticals lies in the tremendous genetic/evolutionary diversity of marine organisms. There are families and entire phyla that are entirely marine.

Marine life provides a unique template for rational drug design. For example, cartilage, an avascular tissue, has been shown to be a powerful source of natural protein inhibitors of capillary growth (angiogenesis) and thereby solid tumor growth. Sharks, for example, are strictly cartilaginous and unique in that they appear to be virtually cancer-free. The purification, characterization and cloning of the gene for short angiogenesis inhibitors from cartilage could result in a new therapeutic agent useful for the treatment of diseases such as cancer, arthritis and many eye diseases. It would also facilitate the design of even more powerful second and third generation inhibitors.

The research for new drugs or biomedical products from marine organisms should be expanded, but needs to be conducted using a rational approach. Particular benefits may accrue if marine biologists search for new products and processes with needed applications unmet by compounds/processes unavilable from terrestrial organisms. This search might include the following approaches:

1) Emphasis on marine groups already known to produce products of application or interest. Examples would include anti-cancer compounds from sponges, tunicates, bryozoans or sharks.

2) Studies of organisms with visible evidence of chemical defenses or unique physiological/molecular processes.

3) Studies of organisms with unique physiologies, such as those that show accumulation of specific toxic metals.

4) Marine toxins that could be coupled with monoclonal antibodies and used as immunotoxins to target specific malignant cells.

Drug development is dependent on an understanding of the mechanism by which a therapeutic agent works. For example, a careful study of a small protein isolated from the horseshoe crab, Limulus polyphemus, shows that it could be a novel therapeutic agent for septicemia (septic shock) and infectious diseases. Its mechanism of action is to bind bacterial endotoxin very tightly thus neutralizing its toxicity -- a mode of action completely different from standard antibiotics. A biologically active recombinant form of this protein has been expressed in yeast.

Once marine-derived potential therapeutics are identified and characterized, second and third generations of these drugs will be developed. For example, antibiotics can be modified to produce more bioactive derivatives; proteins can be cloned, expressed and eventually small, more bioactive synthetic peptide-mimetics will be developed. Structure-function analyses is facilitated by the fact that the marine environment provides an enriched source of these natural products for these studies.

A potential drawback in the development of marine compounds for therapeutic uses is the difficulty in procurement of enough material for clinical evaluations and further practical uses. Many target compounds are found only in minute quantities in rather rare organisms. Because large scale collection of such organisms would create serious problems to the delicate marine ecosystem, it is essential, both for development and preservation, to develop culturable resources that secures the resupply of these important compounds.

Such development of mass culture systems may require the concerted efforts of people with a wide variety of expertise such as biologists, chemists and engineers. Target organisms for pharmaceuticals might include microalgae, bacteria, fungi, tunicates, bryozoa, sponges, etc. The details of their physiology, metabolism, biosynthetic mechanisms are not fully understood, and there are a number of basic questions that must be answered to develop practical production systems. The Japanese government-supported Marine Biotechnology Institute at Kamaishi is already in the process of developing such systems.

Thus, the marine environment provides an unparalleled opportunity to discover and develop new agents for a variety of medical applications from prevention to diagnostics to drug delivery systems and new pharmaceuticals. Sea Grant's Marine Biotechnology program can play a decisive role in stimulating research and development n this important field.

Biomaterials for Sensor and Probe Development

The need for measurements is pervasive. Measurements are important for environmental monitoring, clinical monitoring, and industrial process control. In the marine science community measurements are important for understanding the oceans. Materials isolated from biological sources can be employed extensively to solve a variety of measurement needs. There are two major problems that can be addressed using such biomaterials. The first is to solve the measurement requirements of the marine scientific community. This problem can be addressed by bringing biomaterials from either conventional or marine sources to solve particular measurement needs in the ocean environment. The second problem is to identify biomaterials from marine sources that are useful for making measurements both within and outside the marine sciences. In this case, biomaterials derived from marine sources can be employed to solve a variety of measurement needs. Each of these problems will be addressed below.

Measurement Requirements of Marine Scientific Community
The needs of the marine scientific community are wide ranging:

Carbon Cycle
In order to understand the carbon cycle a variety of analytes must be measured. These include nutrients, carbon in its various forms and a variety of physical and other chemical parameters. Understanding the biogeochemical cycle has environmental importance variety of global warming scenarios.

Biochemical Factors that Control Productivity
These parameters can be obtained by direct measurements of important sea water constituents (e.g., nutrients and metal ions). In situ measurements may also be used for sea-truthing of space-based measurements of ocean color. Measuring net fluxes of nutrients and other components of the food chain is an interest as well.

Aquaculture/Fisheries
Monitoring aquaculture quality control and optimizing aquaculture may require a variety of measurements. These include nutrient monitoring, accurate detection of pathogenic bacteria and viruses, oxygen monitors, as well as other physical and chemical parameters. Sensor systems are also necessary for monitoring chemicals and micro-organisms of importance for the use of wild and aquaculture organisms as food. These systems will address concerns of safety and product quality.

Environmental Quality
Environmental measurements are important for a variety of reasons. Measurements of anthropogenic contaminants resulting from form a variety of sources including, atmospheric fallout, industrial effluents, and oil spills are important for understanding the effect of such contamination on water quality and ecosystem health. A second source of concern is in the area of natural products measurements such as paralytic shell fish toxins (PSP), domoic acid, which causes amnesic shellfish poisoning (ASP), and other products of algal blooms. Geological sources such as oil seeps and thermal vents may also be of interest.

Biodiversity
With recent interest from the Department of Interior to perform an extensive biological survey, it is equally important to assess the biodiversity of the marine environment. Biodiversity is a three step process involving assessment, screening for unknowns, followed by management of the resource. Intelligent management decisions can only be accomplished with full information.

Analysis Techniques
There are three distinct steps for performing an analysis - sampling, pretreatment and measurement. Sampling and pretreatment invariably introduce bias into the measurement. Therefore, any measurement scheme should strive to reduce such techniques. The optimal approach is to make in situ measurements, which are performed either continuously, using devices called sensors, or periodically employing sensors or probes. In situ measurements, using either sensors or probes, reduce the risk posed by sampling and typically do not require any pretreatment of the sample. Probes differ from sensors only in that they can be used to make a single measurement and must be recharged before subsequent measurements can be made. Sensors are either continuous or periodic depending on the frequency of measurement necessary, available power sources, and sensor lifetime.

The other approach to analysis is use of sampling techniques. When sampling techniques are required it is preferred that immediate analysis be performed to minimize the difficulties associated with storage. Immediate analysis can be performed with analytical instrumentation that resides in the water and periodically samples and performs the analysis. When such techniques are not available delayed analysis is required. In these cases either preserved or incubated samples must be obtained for subsequent analysis. These techniques are the least desirable and pose the greatest chance for contamination or sample perturbation.

Biomaterials for Sensor and Probe Development
To expand the ability of marine scientists to make measurements, a variety of biotechnological techniques utilizing biomaterials are possible. There are two broad areas where biomaterials can have an impact. First, using biomaterials to expand the use of bioassays in ocean sciences. For example, the use of tissues, cell cultures, and whole animal bioassays to assess the status of a particular sample. Second, selective recognition molecules can be used such as antibodies and other immune recognition molecules, nucleic acids, enzymes, receptors, carbohydrates, and other biological acceptor molecules to enhance the recognition chemistry available for a variety of analytical measurements.

In addition to using biomaterials to solve ocean measurement needs there is a great opportunity to identify biomaterials from marine sources that are useful for making measurements. Marine-derived biomaterials are a potentially huge resource due to the large biodiversity of marine organisms. In addition, marine organisms possess unique physiologies due to their unique environment. For example, hydrothermal vents have extremely high temperatures and organisms that reside near such vents have acquired an unusual stability to such high temperatures. Bottom-dwelling organisms possess stability at extremely high pressures. Most marine animals operate at relatively low temperature. Adaptation to extreme environments has occurred through the development of biological macromolecules with novel properties that might serve a variety of measurement needs.

Recognition Systems
There are three classes of biomaterials that might be useful for solving sensing and measurement needs. First is the area of new recognition systems. One of the major challenges confronting sensor and other assay development is the identification of new recognition species. Such recognition species include the same types of materials described above. For bioassays tissue, cell culture and whole animals all offer the potential for analyzing a wide variety of biologically- active materials including nutrients, toxins and materials of potential medical importance. A different class of recognition system are binding species such as antibodies or other immune recognition species, nucleic acid based systems, enzymes, carbohydrates, and receptors. These materials can all be coupled to sensors and/or other diagnostic devices that enable selectivity to be achieved. With the unique metabolism and biochemical pathways exhibited by many marine organisms the promise exists for unique selectivities to be isolated. Furthermore, as described above, the extreme environments of hydrothermal vents and the high internal pressures of bottom dwelling organisms should lead to thermally stable, pressure stable, pH stable and high ionic strength-tolerant materials to enhance the longevity and stability of sensors and other diagnostic materials derived from these sources. Many chemical processes require high temperatures and pressures in which such stable recognition species may be employed.

Binding Matrices
A second important class of biomaterials are the binding matrices used for both adhesion and attachment of recognition elements to the transducer. A sensor typically is fabricated by modifying the transducer surface to obtain good adhesion between the recognition layer and the transducer. Many marine organisms have the ability to adhere to a wide range of surfaces. By isolating and using such capabilities, it may be possible to enhance the adhesive interaction between the recognition layer and transducer thereby preventing delamination from occurring. In addition, these matrices tend to have multiple functional groups that make them ideal for binding various recognition species to the polymeric matrix. Such binding matrices may also be useful for cells and organisms employed in bioassays. Enhanced adhesion to culture substrates will enhance the ability of such cultures to grow and be maintained outside of their natural source.

Antifoulants
A third area of use for biomaterials derived from marine organisms is in the area of antifoulants. These antifoulants have many potential applications. For example, sensors used in seawater need to remain clean and free from biofilm formation. Materials put in contact with seawater for long periods of time typically require antifoulants such as tin and copper species that are both toxic and have the potential to interfere with the sensor chemistry. Consequently new materials to coat sensors are needed to maintain a clean interference-free recognition layer. Such materials have a wide variety of environmental applications including groundwater monitoring, seawater monitoring and industrial effluent monitoring. A second major area in which antifoulants are desirable is in the area of invasive medical sensors. Both short and long term fouling may occur including protein absorption, fibrin formation, and platelet aggregation for short term (days) applications and encapsulation for long-term sensing applications. Fibrin and platelet aggregation problems

can be solved by systematically dosing patients with anticoagulant. This approach is undesirable as it leads to increased surgical risk and the possibility of internal bleeding. Consequently, localized treatment of the sensor to prevent such blood and bio-incompatibility would solve a major problem and point the way to long lasting, safe in-dwelling clinical sensors.

Discovering New Biomaterials

There are several methods and approaches to discovering biomaterials from marine organisms. These include directed screening, through such techniques as affinity chromotography to identify unique binding agents, enzyme assays to isolate unique activities, and several immunological methods. A second approach that cannot be minimized in importance is that of serendipity. This approach includes both general serendipity and serendipity based on a piggy back method. For example, during the course of studies on biodiversity a variety of unique materials may be identified.

Advisory and Educational Program Initiatives

The Congressional Act for the marine biotechnology research initiative stated that the funds will be used for supporting research, education and outreach activities. This group met to identify critical issues in education and advisory services for this new strategic research initiative. Secondly, group members explored what opportunities might exist for regional collaborations. These collaborations would work towards the development and submittal of proposal(s) that meet a regional, and where possible, national educational or advisory need.

The marine biotechnology initiative represents a significant focused effort by Sea Grant in a relatively new field. Rapid advances are taking place in biotechnology research and development. This progress is are beginning to have a ripple effect in the marine field, resulting in the application of new techniques and processes to isolate compounds of interest from marine organisms. Scientists are extremely interested in the marine environment because it is a source of unique and new biologically-derived compounds, many of which are elucidated in other sections of this workshop report.

From the perspective of most Sea Grant staff and the public, biotechnology still remains a mystery. However, as Sea Grant embarks on this major new marine biotechnology initiative, efforts must be made to communicate to the public (be they local government officials, industry, or teachers, etc.), the promises and risks associated with marine biotechnology research, development and use.

Biotechnology, and particularly marine biotechnology are relatively new and rapidly advancing fields and an extensive knowledge-base outside of the research community has yet to be developed. Sea Grant can play an important role in educating the public about the potential benefits and risks associated with biotechnology research through its existing network of advisory agents and communications staff. However, before this process can begin, Sea Grant staff must first become knowledgeable themselves.

To date, Sea Grant outreach staff have had little experience in communicating to their constituents the advances taking place in marine biotechnology. The main reason for this is that marine biotechnology research has never played a significant role in Sea Grant-supported research. With the advent of Sea Grant's marine biotechnology initiative, Sea Grant outreach staff must gain a greater understanding of marine biotechnology, from the fundamentals of nomenclature to the most significant advances taking place. Only through having a firm understanding of marine biotechnology will the staff be able to communicate to the public what marine biotechnology research may mean to them. Therefore, the National Sea Grant Program must begin its outreach efforts by first educating its existing outreach staff. Once Sea Grant outreach staff become familiar with the issues of marine biotechnology, they will feel comfortable initiating the outreach and networking activities necessary.

The most effective mechanism to educate Sea Grant staff throughout the nation is the sponsorship of a national intensive workshop in marine biotechnology advances and applications. The workshop will bring together Sea Grant outreach staff to learn firsthand from leading researchers in marine biotechnology and policy, as well as from individuals representing industry. This will allow most of Sea Grant's network of outreach staff to gain the necessary information to begin the process of educating and networking among their constituents.

After the Sea Grant outreach staff has become familiar with marine biotechnology, the next step is to apply that knowledge towards educating the public. Currently there is much mystique around the topic of biotechnology in the public's eye. That mystique must be replaced with knowledge so that the public may make intelligent, well-informed decisions regarding advances in this important field. For example, there is much concern regarding the potential release of genetically enhanced aquaculture species in the open environment. What effect will these altered stocks have on existing wild populations? Should there be strict controls to minimize the potential of an accidental release? Researchers themselves often have difficulty grappling with these questions. Therefore, Sea Grant outreach staff will play an important role in educating the public both about the risks and also the potential benefits from supporting marine biotechnology research.

To effectively and efficiently communicate the latest advances in marine biotechnology, new methods of communication must be combined with more traditional ones. One technique that may be quite effective is the development of short radio 'spots' that focus on a particular aspect of marine biotechnology. These spots would be aired on public radio stations throughout the country and would provide information on the latest results and potential impacts of recent biotechnology developments. This type of approach has been successful for a number of other scientific topics and should be further explored by Sea Grant.

Sea Grant will also need to make outreach efforts to industry. Industry may have the most to gain from Sea Grant-supported marine biotechnology research and must be made aware of the results of such research in a timely and effective manner. This will help to commercialize the results of Sea Grant research, and, through industry comments, ensure that future research accounts for the needs of industry. This may be accomplished through the establishment of an industry consortium for marine biotechnology. MIT's Marine Industry Collegium Program may serve as a model in the development of such a consortium.

Significant, rapid advances are occurring throughout biotechnology; marine biotechnology is only one small, but important subset of that field. For Sea Grant to play an influential role in marine biotechnology research, development and commercialization, it must support the efforts to educate both its outreach staff and the public-at-large. Only then, will Sea Grant-supported research be effectively communicated to fully benefit the public.

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